9

Nutritional value of yoghurt

9.1 Introduction

The chemical composition of a foodstuff provides a useful indication of its potential nutritional value and the data shown in Table 9.1 indicate the main components of some typical natural and fruit yoghurts. If these figures are accepted at face value, then it is evident that yoghurt could prove to be an important introduction to any diet, with the precise impact depending upon the type of yoghurt being consumed. At the same time, it must be accepted that numerical values reveal only part of the story, and even if the almost mystical properties ascribed to yoghurt are ignored for the moment, there are some aspects of the behaviour of yoghurt in the human body that are not revealed by chemical analysis (Robinson, 1977).

It is of some interest, therefore, to look at the constituents of yoghurt in a little more detail and, in particular, to assess the likely nutritional importance of the materials concerned. Earlier studies of the nutritional aspects of yoghurt have been reviewed by Deeth and Tamime (1981) and Alm (1982) and perodically the International Dairy Federation publishes monographs updating the nutritional properties of fermented milks including yoghurt (IDF, 1983a, b, 1988, 1990, 1991, 1992; see also the review by Gurr, 1982). In addition, the following are recommended for further reading on the nutritional properties of yoghurt (Amer and Lammerding, 1983; Renner, 1986; Rao *et al.*, 1986; Rasic, 1987; Bourlioux and Pochart, 1988; Driessen and de Boer, 1989; Biacs and Beczner, 1990; Berner and Lofgren, 1991; Mann, 1993; Khedkar *et al.*, 1993, 1994; Bronzetti, 1994; Gurr, 1987, 1994). Some textbooks (Renner, 1983, 1989; Chandan, 1989) detailing the importance of dairy products in human nutrition together with the proceedings on fermented milks and health which took place at the Netherlands Institute for Dairy Research (NIZO) in the late 1980s (Anon., 1989) deserve a mention.

9.2 Carbohydrates

9.2.1 Available carbohydrates

The expression "available carbohydrates" is intended to cover all those carbon compounds that can be assimilated by the human body and hence can act as a source of energy for metabolism. In the case of natural yoghurt (see Table 9.1), a number of mono- and disaccharides are present in trace amounts, but lactose remains the dominant sugar in natural yoghurt; even after fermentation, the product may contain some 4–5 g 100 g^{-1} lactose (Tamime, 1977; Scrimshaw and Murray, 1988; Barrantes *et al.*, 1994). The reason for this residue is that the process milk is often fortified to 14–16 g 100 g^{-1}TS (total solids) (i.e. up to about 8 g 100 g^{-1} lactose), so that the lactose content of the end product is little different from normal milk. What is different, however, is the effect that these apparently identical levels of lactose can have on people who are so-called lactose-intolerant or lactose maldigestors and the nature of this reaction is of considerable medical interest (Gilliland, 1991).

Lactose intolerance is the inability of humans to metabolise lactose (Rao *et al.*, 1985; Scrimshaw and Murray, 1988; Lerebours *et al.*, 1989; Fernandes and Shahani, 1989a; Dupont and Gendrel, 1992; Alm, 1993). However, most children possess, at birth, the ability to secrete the enzyme lactase (β-galactosidase), so that the lactose in mother's milk is readily broken down into glucose and galactose. These monosaccharides, and especially glucose, are readily metabolised, but as the energy demands of the child increase, so other foods become more important. In many communities this change means that milk plays an increasingly unimportant role in the diet, and as lactose intake falls, so the secretion of lactase declines. A point is then reached quite early in development when lactose can barely be assimilated at all and the free lactose produces a range of unpleasant symptoms, such as abdominal bloating, cramp and diarrhoea. These problems arise through the heterofermentative metabolism of lactose by the natural microflora of the colon, and the gas produced by the coliforms, for example, gives rise to extreme discomfort. This reaction

Table 9.1 Some typical values of the major constituents of milk and yoghurt (all units 100 g^{-1})

Constituent	Milk		Yoghurt[a]			
	Whole	Skim	Full fat	Low fat	Low fat/fruit	Greek-style
Water (g)	87.8	91.1	81.9	84.9	77.0	77.0
Energy value (kcal)	66	33	79	56	90	115
Protein (g)	3.2	3.3	5.7	5.1	4.1	6.4
Fat (g)	3.9	0.1	3.0	0.8	0.7	9.1
Carbohydrate (g)	4.8	5.0	7.8	7.5	17.9	NR
Calcium (mg)	115	120	200	190	150	150
Phosphorus (mg)	92	95	170	160	120	130
Sodium (mg)	55	55	80	83	64	NR
Potassium (mg)	140	150	280	250	210	NR
Zinc (mg)	0.4	0.4	0.7	0.6	0.5	0.5

[a] The nutrient levels in fruit yoghurt will vary with the type of fruit and stabiliser.
NR: Not reported.

Adapted from Holland *et al.* (1991) and Buttriss (1997).

to the ingestion of milk is usually referred to as primary lactose intolerance, and Garza and Scrimshaw (1976) have described a clinical test to confirm this form of deficiency. Similar reactions can, of course, be observed in patients suffering from a congenital absence of lactase, or from walls of the intestine that have become severely disfigured as a consequence of malnutrition but, in the present context, it is the widespread primary intolerance that is most relevant.

Thus, the occurrence of this primary reaction is observed extremely rarely among Europeans who consume milk or processed milk products throughout their lives, but is a common phenomenon in communities where supplies of liquid milk are scarce or erratic. Yet curiously enough, these latter groups may well rely on the production of various types of yoghurt to provide an outlet for any milk which is available, and the failure of lactose in yoghurt (as against lactose in liquid milk) to provoke an intolerance reaction is something of a curiosity.

The most obvious explanations are that either the micro-organisms in the yoghurt continue to metabolise the lactose even after ingestion or the organisms undergo lysis during digestion and the lactase so released ensures that the level of lactose reaching the colon is too low to cause an adverse reaction (Gallagher *et al.*, 1974; Desmaison *et al.*, 1990).

As some strains of *Lactobacillus delbrueckii* subsp. *bulgaricus* are tolerant of low pH, it is feasible to suggest that some breakdown of lactose does continue in the stomach, particularly as the bacteria may be protected, to some extent, within the yoghurt coagulum which can act as a pH buffer. However, a more likely sequence of events is that the low pH of the stomach kills the yoghurt bacteria and that the cell walls of the bacteria then protect the β-galactosidase against the stomach acid. Thus, at pH < 3.0, lactase is rapidly destroyed *in vitro* but, within a bacterial cell, it could pass intact into the alkaline conditions of the intestine (Martini *et al.*, 1987b). Here the bile salts would be expected to lyse the cells (Gilliland and Kim, 1984), so releasing the enzyme into the intestine where it can act on the ingested lactose; some evidence to this effect was found by Goodenough and Kleyn (1976).

One further effect that may be relevant in this context is that yoghurt is already coagulated prior to entering the stomach, while liquid milk is clotted by the acid/enzymes in the body (Davis and Latto, 1957). This difference could mean that the yoghurt coagulum remains partially intact after ingestion, and hence that the lactose remains in the proximity of the disintegrating bacterial cells/escaping lactase (Shah and Jelen, 1991).

Most studies on humans who have been identified as lactose intolerant are in agreement with each other. Such subjects after ingesting live yoghurt had reduced levels of hydrogen secretion in their breath (see Fig. 9.1) and there were fewer reports of diarrhoea or flatulence (Kolars *et al.*, 1984; Savaiano, 1990; Mustapha *et al.*, 1997). Such results indicate that yoghurt, when compared with milk, facilitates the metabolism of lactose due to the intraintestinal digestion of lactose by β-galactosidase released from *Streptococcos thermophilus* and *L. delbrueckii* subsp. *bulgaricus* (Rao *et al.*, 1991). Similar reduced breath hydrogen responses in adult lactose maldigestors were observed when tested against different types of yoghurt (i.e. low or full fat, lactose hydrolysed and frozen) (Martini *et al.*, 1987a; Rosado *et al.*, 1992). The same authors and Onwulata *et al.* (1989) concluded that endogenous lactase originating from the yoghurt micro-organisms is superior to exogenous commercial lactase preparations in alleviating lactose maldigestion. In addition, Pochart *et al.* (1989) have demonstrated that viable yoghurt organisms can, due to the

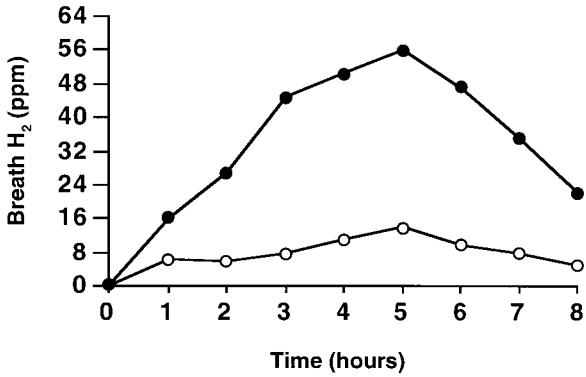

Fig. 9.1 Changes in breath hydrogen for humans ($n = 10$) after ingestion of milk (–●–) or yoghurt (–○–)

Data adapted from Kolars *et al.* (1984).

buffering capacity of the product, reach the duodenum and do have active β-galactosidase. Thus, the starter cultures are to some extent protected from gastric acid secretion and the retention of the microbial lactase inside the cells prevents it from hydrolysing the lactose in the duodenum; mixed yoghurt cultures displayed the greatest β-galactosidase activity, followed by *Lactobacillus acidophilus* and *Bifidobacterium bifidum* (Ordonez and Jeon, 1995).

This degree of physiological acceptability means that yoghurt can provide a useful source of energy in the diet of many consumers, and it is important that while natural yoghurt contains around 6.4 g of carbohydrate $100\,g^{-1}$, fruit yoghurts may contain up to 18–20 g $100\,g^{-1}$ of sucrose and other available carbohydrates (Table 9.1). If each gram of sugar provides around 4 kilo-calories of usable energy, then the contribution of yoghurt towards combating a dietary deficit of carbohydrates can be appreciated, a feature that is enhanced in many brands by the use of modified starch as a stabiliser and at concentrations that may approach 1 g $100\,g^{-1}$ of yoghurt.

Lactic acid is synthesised by the starter culture from lactose which is the principal substrate present in milk (Zourari *et al.*, 1992). Lactic acid occurs in two isomeric forms: L(+) and D(–). In yoghurt, *S. thermophilus* produces the L(+) form, while *L. delbrueckii* subsp. *bulgaricus* releases the D(–) isomer or a racemic mixture DL depending upon the strain. In nutritional terms, the L(+) isomer is the easily digested form and its contribution to the total concentration in yoghurt will vary with the ratio of *S. thermophilus*:*L. delbrueckii* subsp. *bulgaricus*; it is usually between 50 and 70% of the total (Kunath and Kandler, 1980). By contrast, the D(–) isomer is poorly metabolised and an excessive intake is reported to cause acidosis in some children.

9.2.2 Unavailable carbohydrates

Although natural yoghurt is based entirely on milk, stirred fruit yoghurts usually have stabilisers incorporated to reduce whey separation during distribution. The usage of these stabilisers has been considered in detail elsewhere (see Chapter 2),

but it is worth noting that many of them are complex carbohydrates. Thus, guar gum, locust bean gum, as well as the carrageenans and cellulose derivatives are long chain polysaccharides composed of regular arrangements of monosaccharide units and it is significant, in the present context, that the molecules cannot be attacked by digestive enzymes in the human body.

It is for this reason that these hydrocolloidal materials are often referred to as unavailable carbohydrates (Robinson and Khan, 1978) and as such they may contribute to human nutrition by:

- Providing a bulking agent for the contents of the intestine, so stimulating intestinal peristalsis and avoiding some of the risks of colonic malfunction.
- Absorbing some of the potentially toxic chemicals that may be formed in the large intestine as the result of bacterial action.
- Acting to delay the diffusion of sugars to the intestinal wall, a function that could help those prone to postprandial hyperglycaemia. Thus, the surge in insulin production that is required after each meal in order to stabilise the level of glucose in the blood, places an undesirable strain on the hormonal system of even normal subjects and for mild or incipient diabetics, the sudden demand poses particular problems. If the inclusion of unavailable carbohydrates in the meal reduces the rate of entry of glucose into the blood, then the stimulus for insulin production will also decline and this trend towards homeostasis can be regarded as biologically attractive.
- Lowering the cholesterol level in the blood (Jenkins *et al.,* 1975; Roberfroid, 1993).
- Acting in conjunction with the coagulated protein to slow the oro-caecal transit time of lactose, so allowing the microbial lactase to ensure that lactose-intolerant consumers do not suffer discomfort (Marteau *et al.*, 1990).

The level of stabiliser incorporation is, of course, rather low (about 0.5 g in $100\,g^{-1}$), and there is tendency nowadays to avoid their use altogether because some of the plant gums have become expensive and because the less expensive forms often give the product an unacceptable mouthfeel. Nevertheless, some brands of yoghurt do contain gums (Anon., 1990), and Saldamli and Babacan (1996) incorporated sugar-beet fibre into yoghurt at levels of up to $2\,g\,100\,g^{-1}$ without any adverse effect on flavour.

9.3 Protein

The proteins in milk are of excellent quality biologically and both the caseins and whey proteins (α-La and β-Lg) are well endowed with essential amino acids. An indication of levels encountered is shown in Chapter 7 and it is clear that milk is a most valuable dietary component. The fact that the protein content of yoghurt is often elevated by concentration or addition of skimmed milk solids, means that it is an even more attractive source of protein than liquid milk (Table 9.1). The relevance of this point is highlighted by the number of protein-enriched yoghurts that are available in industrialised countries. Consumption of around 200–250 ml of yoghurt per day can easily provide an individual with the minimum daily requirement of animal protein (15 g) (Altschul, 1965; Cheeseman, 1991).

Obviously, such data are impressive in their own right, but two further points

about the protein in yoghurt should be borne in mind. In the first place, it is important that the proteins in yoghurt are totally digestible, a feature enhanced by the fact that some degree of initial proteolysis is caused by the starter organisms themselves. The extent of this breakdown will depend on the strains of bacteria being employed but, in general, at least some release of amino acids and peptides can be expected during incubation and storage (Breslaw and Kleyn, 1973; Butikofer *et al.*, 1995).

The other pertinent characteristic is that the milk proteins in yoghurt are already coagulated prior to ingestion and, in addition to the possible effect discussed earlier, the "soft clot" formed in the stomach may have other benefits. Thus, the contrast between the ingestion of yoghurt and liquid milk has some parallel with the comparative behaviour of warm milk and cold milk for, while the caseins in cold milk form a "hard clot" in the presence of acid in the stomach, the modified caseins (see Chapter 2) in warm milk coagulate more gently (Jay, 1975). The advantages of this latter type of coagulum are alleged to be that the softer structure does not give rise to any feeling of discomfort and that the more "open" nature of the casein aggregates allows the proteolytic enzymes of the alimentary canal freer access during digestion. It is, of course, impossible to quantify, or even assess with any degree of objectivity, effects of this type, but belief in their existence is sufficiently widespread to give some credence to the general hypothesis. What is beyond dispute is that yoghurt is an excellent source of protein and that this fact alone justifies its inclusion in a diet. However, Gaudichon *et al.* (1995) studied the exogenous and endogenous nitrogen flow rate and level of protein hydrolysis in the human (n = 16) jejunum after feeding with ^{15}N-labelled milk and yoghurt and they concluded that: (a) endogenous N secretion was significantly stimulated 20–60 min and 20–40 min after ingestion of yoghurt and milk, respectively, (b) the endogenous N flows over a 4 hour period were similar for milk and yoghurt, whilst the exogenous N flow rates indicated a delayed gastric emptying of yoghurt when compared with milk and (c) the non-protein nitrogen (NPN) flow rate in the jejunum increased significantly after milk and yoghurt due to an increase in the exogenous NPN flow rate, which ranged between 40 and 80%, whilst the net gastrojejunal absorption of exogenous N for milk and yoghurt were similar. It was concluded that the high level of exogenous N hydrolysis reflects the good digestibility of milk and yoghurt; however, fermentation of the milk modifies only the gastric emptying rate of N.

9.4 Lipids

Although much of the yoghurt sold in industrialised countries is produced from skimmed milk, traditional yoghurt has always contained some 3–4 g 100 g^{-1} milk fat (Table 9.1); indeed concentrated yoghurt (labneh) or Greek-style yoghurts will contain 9–10 g 100 g^{-1} fat (Anon., 1997b; Buttriss, 1997). The influence of these lipid materials on the consistency and mouthfeel of yoghurt has been discussed elsewhere, but it should not be forgotten that lipids are an integral part of a balanced diet. Thus, humans have a double requirement for lipids in that they possess:

- storage fat composed of saturated fatty acids and serving as a source of energy or as a protection for vital organs;
- structural fat which, with proteins, forms many of the essential membranes in animal cells, particularly in areas like the brain.

It is essential, therefore, that the human diet provides an adequate source of fats, a point that is of especial relevance for children. Thus, with each gram of fat providing around 9 kcal, fats are a most valuable source of energy. When this figure is viewed in relation to the fact that malnutrition in children is often associated with a lack of calories to metabolise available protein, then the potential relevance of this compact source of energy is evident. It is also important that yoghurt is widely accepted by children as a foodstuff and hence developing countries, in particular, would be well advised to look closely at the merits of yoghurt for school feeding programmes. In addition to this basic advantage of consuming full fat yoghurt, it must also be stressed that milk fat contains an extremely wide range of fatty acids. Most of these are present in the form of various glycerides, but over 400 individual fatty acids have been identified in cow's milk (Patton and Jensen, 1974). Obviously, it is impossible to assign a physiological role to all but a handful of these acids, but the fact that they are present in a normal mammalian secretion merely confirms that ignorance of function should not be equated with no function.

There is, of course, every incentive for a manufacturer to remove the fat from the process milk and sell it as cream, but it is clear that, both organoleptically and nutritionally, the interests of the consumer may be better served by leaving a reasonable level in the end product. Such a proposal would not find universal acceptance, for some authorities would be concerned at the additional intake of saturated fatty acids that would be involved. However, the evidence linking fats of dairy origin with coronary and similar problems is, to say the least, tenuous, and hence yoghurt manufacturers should be encouraged to base their judgements concerning fat content on the broader concept of quality (Gurr, 1992). Whether such an aim is feasible in light of the vociferous anti-cholesterol lobby remains to be seen and, certainly in some countries like the United States, challenging consumer groups could spell financial ruin.

The tragedy of this situation is that it is the consumer who loses out and, once again for no reason capable of objective assessment. The totally irrelevant demand for nutritional labelling of yoghurt and other foods falls into the same category, because it is more than evident that the nutritional value of yoghurt cannot be summarised by a few figures stamped on the side of a retail carton. In effect, therefore, the consumer will be paying for a quite useless set of data, in that the information implies that the designated nutrients will be absorbed into the human body, whereas in fact, chemical analyses should never be equated with nutrient availability and, in the case of yoghurt, any serious consideration of its nutritional value must include the question of whether the product possesses special therapeutic properties. Clearly no label could honestly convey to a consumer that yoghurt may be more than a mere carton of chemical compounds.

9.5 Vitamins and minerals

The increase in solids-not-fat (SNF) in yoghurt as compared with liquid milk carries with it the implication that the level of inorganic ions/unit weight is also going to be higher and this view is confirmed by the data in Table 9.1. In most cases, the figures speak for themselves, but the position of calcium is perhaps, rather special in relation to a typical recommended daily allowance (RDA) of 800 mg (Weaver

and Plawecki, 1994; Anon., 1997a). Thus, not only can yoghurt act as a source of calcium for sufferers of lactose intolerance but, in addition, calcium supplied by yoghurt may be better absorbed and utilised than calcium made available in other forms (Dupuis, 1964; Rasic, 1987); the role of dairy calcium in bone metabolism and prevention of osteoporosis has been recently reviewed by Renner (1994). Phosphorus, magnesium and zinc are also well represented and it is likely that the proportions of the total concentrations available for absorption and utilisation by the body is also high (Buttriss, 1997). However, Galan *et al.* (1991) reported that, under normal conditions, increasing the daily intake of dairy products probably has no effect upon iron absorption from meals already containing appreciable amounts of milk-based components.

Yoghurt contains appreciable quantities of sodium and potassium which may not be suitable for feeding babies less than 6 months (Doyle *et al.* 1981) but, as shown in Chapter 2, the mineral salts in milk can be reduced prior to the production of yoghurt.

The relative availability of vitamins in yoghurt is much more difficult to assess because, unlike minerals, many vitamins are sensitive to the conditions of processing. Thus, the method of fortification, for example, the addition of milk powder or membrane processing, the heat treatment of the milk base, the strains of starter bacteria used and the conditions of fermentation can all alter the concentrations of the more important vitamins (Noh *et al.*, 1994). For this reason, the figures quoted in Table 9.2 should be regarded merely as a guide to the vitamins available in yoghurt, and hence as an indication that, although regarded by many as a convenience food, it is certainly not a trivial item in terms of potential nutritional value. The fortification of yoghurt with vitamins, such as vitamins A or C, is possible (Anon., 1997a, b), and losses over two weeks in storage are unlikely to exceed 50%;

Table 9.2 Some typical vitamin contents of milk and yoghurt (all units $100\,g^{-1}$)

Vitamin	Milk		Yoghurt		
	Whole	Skimmed	Full fat	Low fat	Low fat/fruit
Retinol (μg)	52	1	28	8	10
Carotene (μg)	21	Tr	21	5	4
Thiamin (B_1) (μg)	30	40	60	50	50
Riboflavin (B_2) (μg)	170	170	270	250	210
Pyridoxine (B_6) (μg)	60	60	100	90	80
Cyanocobalamine (B_{12}) (μg)	0.4	0.4	0.2	0.2	0.2
Vitamin C (mg)	1	1	1	1	1
Vitamin D (μg)	0.03	Tr	0.04	0.01	0.01
Vitamin E (μg)	90	Tr	50	10	10
Folic acid (μg)	6	5	18	17	16
Nicotinic acid (μg)	100	100	200	100	100
Pantothenic acid (μg)	350	320	500	450	330
Biotin (μg)	1.9	1.9	2.6	2.9	2.3
Choline (mg)	12.1	4.8	–	0.6	–

Tr: Trace.

Adapted from Deeth and Tamime (1981) and Holland *et al.* (1991).

since low fat yoghurt is very popular in many countries, fortification with vitamin A should become mandatory in order to maintain the nutritive value of milk.

Some relevant aspects of the vitamin content of yoghurt have been reported by Rao *et al.* (1984) and Rao and Shahani (1987). It is of interest that certain B group vitamins are synthesised by the starter cultures. Kneifel *et al.* (1989) monitored these vitamins in yoghurt during fermentation using eight commercially available cultures and they concluded that using short time (i.e. 3–4 hours) incubation at 42°C, the starter cultures enriched the vitamins during fermentation by more than 20%, for example thiamin (two cultures), pyridoxine (four cultures), folic acid (one culture) and biotin (two cultures). Only two starter cultures were used to compare vitamin profiles at different incubation temperatures, but it was observed that fermenting the milk at 30°C for 14–16 hours led to a lower production of folic acid, but an increased concentration of thiamin and nicotinic acid. Therefore, it is important to use selected strains of the yoghurt starter cultures and processing conditions in order to maintain the nutritional properties of yoghurt.

9.6 Yoghurt and health

Although yoghurt and similar foods have long occupied a place in the diets of peoples from the Middle East and central Europe, the western world adopted a totally casual attitude to the product until rumours of its health giving properties became rife. In particular, the views of Metchnikoff (1910) linking longevity among the hill tribes of Bulgaria with their consumption of yoghurt caused a considerable flurry of interest.

In essence, it was suggested that one aspect of approaching senility in humans involved an undesirable passage of noxious compounds from the intestine to the blood stream and that these chemicals arose from the action of putrefactive bacteria in the lower ileum and colon. If the activity of these bacteria could be suppressed, then, so it was argued, the adverse effects of their metabolic products would no longer be manifest and the individual might anticipate a longer and healthier life. Such an hypothesis sounded perfectly reasonable, and the role of yoghurt in curtailing the putrefactive bacterial action was readily explained as follows. First, the lactic acid bacteria in yoghurt are tolerant of a low pH, whereas most bacteria show optimum growth and metabolism around neutrality. Therefore, as the acidic yoghurt passed along the intestine, the lactic acid in the food and, perhaps, that still being secreted by the bacteria, would kill the undesirable microflora. Second, it was further suggested that this effect of the yoghurt was enhanced by the ability of *L. delbrueckii* subsp. *bulgaricus* to become established in the intestine, and gradually to dominate the resident microflora. This latter change ensured the continued absence of the putrefactive organisms even during periods of reduced yoghurt availability and hence the vitality of the consumer would be maintained.

At present, the consensus among scientists is that the yoghurt bacterial cultures (*S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*) are unable to adhere to the mucosal surfaces of the intestinal tract although, in some cases, there are conflicting results which could be attributed to strain variation, differences in experimental design and/or the results of animal studies being wrongly applied to humans.

Over the years these original ideas have been the subject of intense discussion and investigation and it has become clear that the critical factor is the microflora of

the product (Tomar and Prasad, 1989; Lascar, 1995). Thus, while yoghurt should have a microflora consisting of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* alone (Bourlioux, 1986; FAO/WHO, 1990), the more recent entrants into the market may contain *S. thermophilus*, *L. acidophilus*, *Lactobacillus paracasei* subsp. *paracasei* and/or *Bifidobacterium* spp. These latter products, often referred to as bio-yoghurts, may be similar to yoghurt in terms of chemical composition, but the impact of the microflora on the digestive system of the consumer is totally different. For this reason, the health implications of consuming these products will be dealt with separately.

9.6.1 Therapeutic properties of yoghurt

There is no doubt that bacteria in the large intestine produce a range of phenolic compounds, such as sketol and indole, which could damage living tissue. Whether they could have any discernible effect on the intestinal wall, or even be absorbed, will depend on their concentration, the ability of other gut contents (e.g. hydrocolloids) to absorb them and their residence time, but, nevertheless, there is definite concern over their possible involvement in the initiation of cancer in the lower intestine (Aries *et al.,* 1969; Sellars, 1991). Any process that tended to suppress their production could, therefore, be advantageous, and the action of lactic acid in inhibiting the growth/metabolism of the putrefactive bacteria could be one such process.

Whether, in fact, any of the acid in yoghurt can survive the neutralising effect of the bile components is open to debate, but the prospect remains that yoghurt could change, albeit slightly, the pH gradient within the intestine. If this change does occur, then there could well be a basis of truth in Metchnikoff's proposal, and certainly the traditional products of Bulgaria would have been extremely acidic (see also Friend *et al*., 1983; Hitchins and McDonough, 1989; Fernandes and Shahani, 1989b, 1990; Reid *et al*., 1990; Kotz *et al*., 1990; Marteau *et al*., 1993; Lin, 1995).

A hypocholesterolaemic action has also been attributed to yoghurt (Mann and Spoerry, 1974; Mann, 1977; Hepner *et al.,* 1979). The exact reason for this effect is not clear (Richardson, 1978), but the fact that yoghurt is more active in this respect than unfermented milk implies that some enzyme system or biochemical compound of bacterial origin may well be involved. Hydroxymethyl glutarate has been proposed as one metabolite of starter cultures that could limit cholesterol synthesis but, for the present, both the reality of the phenomenon and its possible cause remain subjects for speculation (Anon., 1987).

It has been noted in studies with rats and mice that the consumption of yoghurt, live or pasteurised, inhibited the growth of certain types of tumour, and it has been suggested that some factor in the cell walls of the bacteria could be responsible for the effect (Gilliland, 1991). Whether such results can be interpreted as applicable to humans is another matter, but it is a possible benefit of yoghurt consumption that cannot be dismissed (Morissette *et al.*, 1991).

Similarly, stimulation of the normal microflora of the gut has been attributed to the regular consumption of yoghurt and it is proposed that the lysing cells of the starter bacteria release vitamins or other growth factors that encourage the development of *L. acidophilus*, for example, in the small intestine (Robinson, 1989). Some clinical evidence does exist to support this idea but, as with many human studies, it is difficult to establish how widespread the impact of regular consumption would be in a normal population of consumers from a given community.

Table 9.3 Update of current studies of health promoting aspects of yoghurt and related products[a]

Test model or subject	Comments	References
Humans (n = 194)	Elderly patients (males and females ~72 years old) fed a mixture of prune whip and yoghurt improved the bowel movement against constipation and only very few required laxative.	Ferrer and Boyd (1955)
Humans	Lactinex®, a pharmaceutical preparation of *L. acidophilus* and *L. delbrueckii* subsp. *bulgaricus*, ingested for 1 week did not reduce the incidence of traveller's diarrhoea, whilst Gotz *et al*. (1979) reported that the same product was effective in preventing ampicillin-induced diarrhoea.	Pozo-Olano *et al*.(1978)
Humans	Yoghurt and Ca^{2+} supplementation of the diet altered cholesterol metabolism in females (n = 16), but not in males (n = 5).	Bazzarre *et al*. (1983)
Humans and *in vitro*	Survival of the yoghurt organisms in human stomachs and adhesion to intestinal cells was much lower when compared with *L. acidophilus*; by careful strain selection it is feasible to achieve elevated levels of *Lactobacillus* spp. in the intestine.	Conway *et al*. (1987)
Children (n = 156)	Ayran (a Turkish drinking yoghurt) was used successfully to dissolve oral rehydration salts in the treatment of diarrhoea in children aged 3–48 months.	Caglayan *et al*. (1989)
Men (n = 18)	Eating yoghurt had no effect on plasma cholesterol levels in normolipidemic in males.	McNamara *et al*. (1989)
Children (n = 52)	Children aged 3–36 months with persistent diarrhoea were fed yoghurt or milk, and the results suggest a clinical advantage of feeding yoghurt.	Boudraa *et al*. (1990)
Humans (n = 68)	Chronic high level consumption of live yoghurt (450 g day^{-1} for 4 months) showed the following results: (a) no negative side effects were found in many parameters studied including cholesterol, (b) significant and potential increase in serum ionised Ca^{2+} levels, and (c) increased production of γ-interferon isolated T cells.	Halpern *et al*. (1991)
Women (n = 13)	Daily ingestion of yoghurt (~230 g for 6 months) containing *L. acidophilus* decreased both candidal vaginitis colonisation and infection.	Hilton *et al*. (1991)
Women (n = 32)	Female patients with bacterial vaginosis were treated by intravaginal application with yoghurt and the results were favourable, because the continuous adjustment of the vagina pH and implantation of lactobacilli flora are crucial in normal vagina ecology.	Neri *et al*. (1993)
Boys (n = 9) & girls (n = 11)	Five out of six lactose maldigestors had decreased symptoms and significant reduction in breath H_2 excretion following eating yoghurt.	Montes *et al*. (1995)
Humans (n = 259)	Results of questionnaire survey do not support the hypothesis of an increased consumption of fermented milks or dietary calcium decreases the risk of colon cancer.	Kampman *et al*. (1994a)

Table 9.3 *Continued*

Test model or subject	Comments	References
Men (n = 331) & Women (n = 350)	Studies conducted in USA suggest that: (a) total milk and fermented dairy products consumption did not relate to colorectal adenoma risk and (b) vitamin D from supplements rather than diet was slightly and universely associated with such risk among women only.	Kampman *et al*. (1994b)
Children (n = 49)	Different preparations of lactic acid bacteria and *Lactobacillus* GG [currently known as *L. rhamnosus* (Tamime and Marshall, 1997)] which were fed to children, and not the yoghurt starter cultures have promoted serum and intestinal response to rotavirus.	Majamaa *et al*. (1995)
Mice	Animals implanted with Ehrlich ascites tumour and fed with yoghurt showed inhibition of these cells suggesting that the antitumour factor(s) is synthesised by the starter culture.	Friend *et al*. (1982), Friend and Shahani (1984)
Rats	Both starter cultures failed to colonise the gut of germ free rats maintained on stock diet and yoghurt was administered orally; feeding of yoghurt altered the lactobacilli flora of the gut from predominantly *Lactobacillus reuteri* to *Lactobacillus salivarius*.	Garvie *et al*. (1984)
Mice (n = 40)	In obese mice, the hepatic lipid was significantly greater in the yoghurt (Y) diet than in the same product supplemented with dietary chromium (Y + Cr), whilst the plasma immunoreative insulin level was lower in animals fed Y + Cr which was significantly correlated with hepatic lipid and plasma cholesterol.	Li and Stoecker (1986)
Rats (n = 10)	Apparent protein digestion (*in vivo*) in rats was higher in the yoghurt diet.	Lee *et al*. (1988)
Rats (n = 36 & 20)	Yoghurt fed rats showed a significant lower incidence of gastric tumour (50%) when compared with the controls.	Morishita and Shiromizu (1990)
Rats (n = 40) & humans (n = 133 & 289)	Results *may* provide protection against tumour development, possibly via stimulation of the immune system.	Schaafsma *et al*. (1990)
Mice (n = 20)	Milk fermented with *L. delbrueckii* subsp. *bulgaricus* showed no effect on the humoral immune response when fed to mice, but a significant increase in the broncho-alveolar IgA level after 8 days.	Moineau and Goulet (1991a)

Table 9.3 *Continued*

Test model or subject	Comments	References
Mice ($n = 24$)	No stimulatory effect on the phagocytic activity of pulmary alveolar macrophages (Amø) in mice was observed after administering fermented milk made with lactococci and *S. thermophilus* + *L. delbrueckii* subsp. *bulgaricus*; the results from this work and Moineau and Goulet (1991a) suggest that the proteolytic activity of fermented milks might be implicated in the stimulation of non-specific immune system in mice rather than the degree of proteolysis.	Moineau and Goulet (1991b)
Hamsters ($n = 10$)	Yoghurt did not exhibit any bactericidal activity in the prevention of *Clostridium difficile* infection in hamsters.	Kotz *et al*. (1992)
Mice ($n = 5$)	Groups of mice were fed Deodan® [cell wall product of *L. delbrueckii* subsp. *bulgaricus* (i.e. patented by I. Bogdanov, strain tumoronecroticance B51-ATCC 218165)], which is a primer and trigger of endogenous tumour necrosis factor-α (TNFα), is useful for the treatment of neoplastic disease in humans.	Davidkova *et al*. (1992)
Mice ($n = 10$)	Mice fed with yoghurt (unheated or heated post fermentation) or milk fermented with *L. paracasei* subsp. *paracasei* LcFM exhibited higher kinetics of specific antibody responses when compared with the control (i.e. milk fed mice); the IgG_{2a} levels remained stable, but the results suggest that fermented milks stimulate the systematic immune system.	Portier *et al*. (1993)
Mice	Deodan® (see above) fed to mice activated the phagocytic secretory functions of mononuclear cells and increased the host resistance to bacterial infection.	Popova *et al*. (1993)
Rats ($n = 8$)	Feeding yoghurt or high Ca^{2+} milk enhanced the resistance to *Salmonella enteritidis* infection by lowering the luminal cytolytic activity or diminishing the Fe^{2+} availability for the pathogen to grow.	Bovee-Oudenhoven *et al*. (1996)
Rats ($n = 54$)	Ordinary yoghurt had no hypocholesterolaemic effect, but the same product made with lactose hydrolysed WPC[b] and fermented milk with *B. bifidum* lowered the serum cholesterol level in the blood.	Beena and Prasad (1997)

[a] Health studies on fermented milks made with *L. acidophilus* or *Bifidobacterium* spp. are not included. [b] Whey protein concentrate.

n is the number of subjects in the study.

Up to the late 1980s most of the nutritional studies using humans or animals have been reviewed extensively by the International Dairy Federation Group F20 (IDF, 1991), and they concluded the following:

- Some data reported in the literature are not based on well designed experiments and not all the interpretations given are based on differences that are statistically significant.
- *In vitro* results cannot be found always *in vivo* and observations found in animals cannot be translated directly to humans.
- There are problems in generalising the results given the large number of types of micro-organism used.

However, substantial progress in knowledge of this subject has been reported and in an effort to update the data published by IDF (1991), Table 9.3 summarises some of the nutritional studies since the late 1980s (see also Hargrove and Alford, 1978, 1980) relating to yoghurt and other fermented milks.

9.6.2 Therapeutic properties of bio-yoghurt

It is well known that *S. thermophilus* is intolerant of acidity and hence few cells of this species will survive passage through the stomach; even *L. delbrueckii* subsp. *bulgaricus*, which is able to resist acidity to a much greater degree, is unlikely to reach the intestine in a viable state (Accott and Labuza, 1972). The resistance of these same bacteria to bile salts, including sodium taurocholate and glycolate, is also poor (Lembke, 1963), even though certain strains of *L. delbrueckii* subsp. *bulgaricus* have been implanted in the intestines of laboratory rats (Mabbit, 1977). Consequently, the general consensus is that neither *S. thermophilus* nor *L. delbrueckii* subsp. *bulgaricus* survive the digestive process in humans.

However, the new generation of so-called bio-yoghurts has a very different microflora from the traditional product, and indeed the usual absence of *L. delbrueckii* subsp. *bulgaricus* from bio-products has led to some debate about whether it is appropriate to use the term yoghurt at all (see later). Thus, in bio-yoghurts the usual flora may include *L. acidophilus, L. paracasei subsp. paracasei* or *Lactobacillus paracasei* biovar *shirota*, *L. rhamnosus, L. reuteri, Lactobacillus gasseri*, *Bifidobacterium adolescentis, B. bifidum, Bifidobacterium breve*, *Bifidobacterium infantis* and *Bifidobacterium longum*, and more recently *Bifidobacterium lactis* (Mitsouka, 1990; Romond and Romond, 1990; Speck *et al.*, 1993; Pedrosa *et al.*, 1995; Anon., 1996; Marshall and Tamime, 1997). In addition, some products contain *Bifidobacterium animalis*; this latter species is attractive for the manufacturer in that it grows more rapidly in milk than the other species of *Bifidobacterium* mentioned above but, unlike the other species, it has never been isolated from the human intestine. Certain *in vitro* studies are reported to show that strains of *B. animalis* can attach to epithelial cells of human origin but, even so, the use of the species in products alleged to have health-promoting properties is the subject of some debate.

What is important about this group is that all the species are natural inhabitants of the human intestine, unlike *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, with the lactobacilli colonising the distal portion of the small intestine and the bifidobacteria forming one of the dominant groups in the colon. In the small intestine, the lactobacilli occupy both the lumen of the gut and physically attach to specific receptors on the epithelial cells (Salminen *et al.*, 1993). In this niche, the

microflora, which will be composed of a number of species of *Lactobacillus*, occupy the surface area of the intestine, absorb nutrients, secrete lactic acid and, perhaps, antimicrobial compounds (Shahani *et al.*, 1976; Barefoot and Klaenhammer, 1983; Tamime and Marshall, 1997). One prophylactic effect of this combination of activities is that bacteria capable of causing intestinal infections cannot compete, and hence an active population of lactobacilli can provide a degree of protection against *Salmonella* spp. and other causes of traveller's diarrhoea (Alm, 1991; Marteau and Rambaud, 1996). The same high population of lactobacilli will metabolise lactose, so ensuring that the concentration of any residual sugar reaching the colon is sufficiently low to avoid adverse symptoms.

These two results of colonisation of the small intestine by lactobacilli are well established, but Sellars (1989, 1991) has raised the possibility that the same population may also stimulate the immune system of the body, lower serum cholesterol levels and offer some protection against certain forms of cancer (for further information refer to Perdigon *et al.*, 1986, 1990, 1991, 1994, 1995a, b). Some evidence in favour of these ideas does exist, but it is important to note that the results suggest that variation between individuals is critical for success or failure. In other words, it is likely that the intestinal lactobacilli are able to protect most people from food-borne infections, but the other benefits may only be manifest in a small fraction of any given population.

Consequently, great care must be exercised in statements about the health-promoting effects of bio-yoghurts, and Table 9.4 attempts to highlight those influences of ingested cultures that are widely accepted as valid. Some of the other possible advantages mentioned in Table 9.3 may be apparent in some people but, at the present time, the evidence is not convincing, a point borne out by the health claims that are currently permitted on bio-products, for example: "This product contains a culture which may improve the health of the digestive tract". Any further claim(s) would almost certainly be deemed to be misleading by any responsible advertising standards authority.

The bifidobacteria, by contrast, occupy the lumen of the colon and, more specifically, colonise the walls in very high numbers. In this zone, the various species of *Bifidobacterium* occupy the surface area of the intestine, absorb nutrients, secrete lactic and acetic acid and, perhaps, antimicrobial compounds (Gibson and Wang, 1994). However, the dominance of bifidobacteria at the wall of the colon is enhanced by the ability of the genus to metabolise mucin, a complex polysaccharide that eases the passage of faeces (Robinson and Samona, 1992). The population is able, therefore, to prevent colonisation of the walls of the colon by undesirable bacteria (e.g. *Escherichia coli*) or yeasts (e.g. *Candida* spp.) and so protect the individual from diarrhoea associated with overgrowth by yeasts or coliforms. Suppression of the growth of putrefactive bacteria in the faeces is a further advantage deriving from the presence of an active population of bifidobacteria, and it is proposed that this restriction could lower the risk of carcinogenic compounds being liberated during fermentation in the colon (Gotti, 1977; Grill *et al.*, 1995; Rowland, 1996).

Whatever the final outcome of the various controversies surrounding the precise role of the major components of the intestinal microflora, there is no doubt that they are essential for the healthy functioning of the intestine and the population levels of both the lactobacilli and bifidobacteria can be reduced dramatically by outside influences. The administration of antibiotics by the oral route is one obvious adverse factor (Colombel *et al.*, 1987), but radiation and disease can prove equally

Table 9.4 Some of health-promoting activities attributed to cultures in yoghurt and bio-yoghurt and an indication of their likely validity for humans

Action/effect	Alleged health benefit	Established in humans[a,b]
In digestive tract	Active against *Helicobacter pylori*	
	Enhanced lactose digestion	✓
	Stimulation of intestinal immunity	
	Stabilisation of Crohn's disease	
	Stimulation of intestinal peristalsis	
On intestinal microflora	Improves balance between microbial populations	Increase in faecal bifidobacteria
	Decrease in faecal enzyme activity	✓
	Colonisation of intestinal tract	✓
	Reduced carrier time for *Salmonella* spp.	
On diarrhoea	Prevention/treatment of acute diarrhoea	✓
	Prevention/treatment of rotavirus diarrhoea	✓
	Prevention of antibiotic-induced diarrhoea	✓
Other effects	Improved immunity to disease	
	Suppression of some cancers	
	Reduction in serum cholesterol	
	Reduction in hypertension	

[a] More than one publication and no conflicting evidence. [b] A tick indicates confirmed treatments in humans.

After Sanders (1994) and Saloff-Coste (1997).

destructive (Robinson and Samona, 1992). Even alcohol or strong foods like onions or garlic can damage the microflora of some people and it is interesting to speculate whether individuals within specific populations acquire microflora that are tolerant of the major food items in a typical diet.

9.7 Conclusion

The general view is that citizens of western societies are at some risk of suffering damage to their intestinal microflora and hence the question has arisen; could a damage situation be alleviated by the consumption of bio-yoghurt? In general, it is now agreed that bio-yoghurts can have a positive therapeutic effect (Sellars, 1991; Tamime *et al.*, 1995), provided that:

- The product contains at least 1.0×10^6 viable cells of *Lactobacillus* and/or *Bifidobacterium* ml^{-1} as consumed. In the U.K., the majority of bio-yoghurts appear to meet this requirement by a considerable margin (Anon., 1997c), but reports from elsewhere have been more variable (Rybka and Kailasapathy, 1995; Shah *et al.*, 1995).
- The organism is of human origin, so that not only will it withstand transit through the stomach and upper digestive tract, but it will be able to colonise/become implanted upon the epithelial walls of the lower intestine.
- Consumption is on a regular basis – perhaps 200–300 ml per week, with the exact definition of regular depending upon the individual and his/her life style.

If these provisions are met, then there can be little doubt that the cultures in bio-yoghurts can be described as oral probiotics, that is, "living micro-organisms which, upon digestion in certain numbers, exert health benefits beyond inherent basic nutrition" (Buttriss, 1997). Whether the cultures act by replacing severely damaged native microflora or merely enhance recovery of a depleted population will depend upon the specific circumstance, but there is growing evidence that the effect is real enough.

If the consumer appeal of both natural and fruit yoghurts/bio-yoghurts is also placed on record, together with their excellent performance in respect of public health, then it is evident why the market for these products is an expanding one.

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